abcam

Product datasheet

Anti-TPH2 antibody [EPR25100-29] ab288067

Recombinant RabMAb

1 References 17 Images

Overview

Product name Anti-TPH2 antibody [EPR25100-29]

Description Rabbit monoclonal [EPR25100-29] to TPH2

Host species Rabbit

Specificity No suitable positive material of human species is available for in-house IHC-P QC, and it worked

well on overexpressed cells pellets of human species.

Suitable for: WB, IHC-P, IHC-Fr, IP, Flow Cyt, ICC/IF **Tested applications**

Reacts with: Mouse, Rat, Human Species reactivity

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Human brain, Mouse midbrain, Rat midbrain, HEK-293T transfected with Tryptophan 5-

> hydroxylase 2 expression vector with myc-His-tag® whole lysates. IHC-P: Mouse midbrain, Mouse cerebrum, Rat midbrain, Rat cerebrum tissues. IHC-Fr: Mouse midbrain, Rat midbrain tissues. ICC/IF: 293T+OE-1169 cells. Flow Cyt: HEK-293T transfected with a human Tryptophan 5hydroxylase 2 vector with myc-His tag. IP: HEK-293T transfected with Tryptophan 5-hydroxylase 2

vector with myc-His-tag®, Mouse midbrain cells.

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EPR25100-29

Isotype IgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab288067 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/1000. Predicted molecular weight: 56 kDa.
IHC-P		1/2000.
IHC-Fr		1/500.
IP		1/30.
Flow Cyt		1/500.
ICC/IF		1/500.

Target

Tissue specificity Brain specific.

Pathway Aromatic compound metabolism; serotonin biosynthesis; serotonin from L-tryptophan: step 1/2.

Involvement in disease Genetic variation in TPH2 may influence susceptibility to major depressive disorder (MDD)

[MIM:608516].

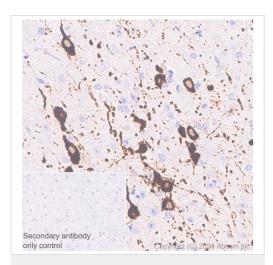
Defects in TPH2 are the cause of susceptibility to attention deficit-hyperactivity disorder type 7 (ADHD7) [MIM:613003]. ADHD is a neurobehavioral developmental disorder and is primarily characterized by the co-existence of attentional problems and hyperactivity, with each behavior occurring infrequently alone. Note=Naturally occurring variants of TPH2 with impaired enzyme activity could cause deficiency of serotonin production and result in an increased risk of

developing behavioral disorders.

Sequence similaritiesBelongs to the biopterin-dependent aromatic amino acid hydroxylase family.

Contains 1 ACT domain.

Images

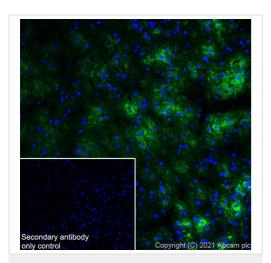


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded Mouse midbrain tissue labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in mouse midbrain. The section was incubated with ab288067 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) .

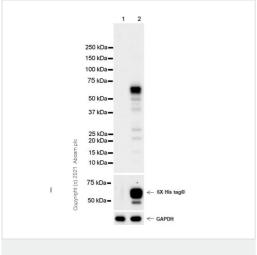
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Frozen sections) - Anti-TPH2 antibody [EPR25100-29] (ab288067)

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Rat midbrain (fresh) tissue labeling TPH2 with ab288067 at 1/500 (1.26 ug/ml) dilution followed by **ab150081**Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/mL) dilution (Green). Positive staining on rat midbrain is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) preadsorbed at 1/1000 (2 ug/mL) dilution.



Western blot - Anti-TPH2 antibody [EPR25100-29] (ab288067)

All lanes : Anti-TPH2 antibody [EPR25100-29] (ab288067) at 1/1000 dilution

Lane 1 : HEK-293T (human embryonic kidney) transfected with an empty vector (vector control), containi a myc-His-tag®, whole cell lysate

Lane 2: HEK-293T transfected with Tryptophan 5-hydroxylase 2 expression vector containi a myc-His-tag®, whole cell lysate

Lysates/proteins at 10 µg per lane.

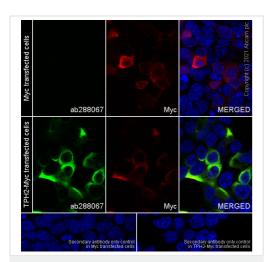
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution

Predicted band size: 56 kDa

Blocking and diluting buffer and concentration: $5\% \ NFDM/TBST$

Exposure time: 10 seconds



Immunocytochemistry/ Immunofluorescence - Anti-TPH2 antibody [EPR25100-29] (ab288067)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized 293T+OE-1169 cells labelling TPH2 with ab288067 at 1/500 (1.26 ug/ml) dilution, followed by **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody ready to use(Green). Confocal image showing cytoplasmic staining in 293T cells transfected with myc-tagged TPH2 expression vector. is observed. Myc-Tag Mouse mAb (Alexa Fluor® 647) was used to counterstain tubulin at 1/100 (0.38ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) is ready to use.



Immunoprecipitation - Anti-TPH2 antibody [EPR25100-29] (ab288067)

TPH2 was immunoprecipitated from 0.35 mg HEK-293T transfected with Tryptophan 5-hydroxylase 2 expression vector containing a myc-His-tag® whole cell lysate 10 ug with ab288067 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab288067 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

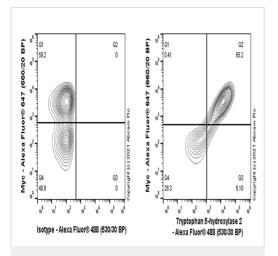
Lane 1: HEK-293T transfected with Tryptophan 5-hydroxylase 2 expression vector containing a myc-His-tag® whole cell lysate 10 ug

Lane 2: ab288067 IP in HEK-293T transfected with Tryptophan 5-hydroxylase 2 expression vector containing a myc-His-tag® whole cell lysate

Lane 3:Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab288067 in HEK-293T transfected with Tryptophan 5-hydroxylase 2 expression vector containing a myc-His-tag® whole cell lysate

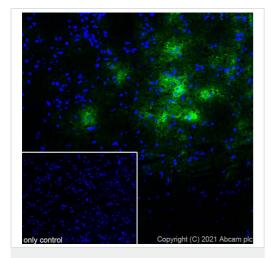
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second



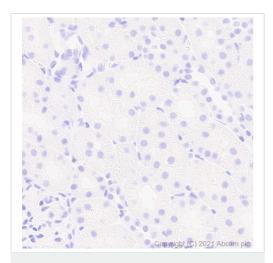
Flow Cytometry - Anti-TPH2 antibody [EPR25100-29] (ab288067)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HEK-293T (Human embryonic kidney epithelial cell) transfected with a human Tryptophan 5-hydroxylase 2 expression vector containing a myc-His tag cells labelling TPH2 with ab288067 at 1/500 dilution (0.1ug)(Right) compared with a Rabbit monoclonal IgG (ab172730) (Left) isotype control. A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



Immunohistochemistry (Frozen sections) - Anti-TPH2 antibody [EPR25100-29] (ab288067)

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Mouse midbrain (fresh) tissue labeling TPH2 with ab288067 at 1/500 (1.26 ug/ml) dilution followed by **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed at 1/1000 (2 ug/mL) dilution (Green). Positive staining on mouse midbrain is observed. The nuclear counterstain was DAPI (Blue). Secondary antibody control: Secondary antibody is **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed at 1/1000 (2 ug/mL) dilution.

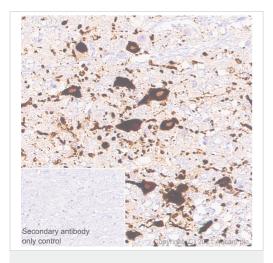


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded Rat kidney tissue labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). No staining in rat kidney. The section was incubated with ab288067 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) .

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

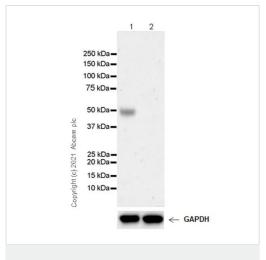


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded Rat midbrain tissue labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in rat midbrain.The section was incubated with ab288067 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Western blot - Anti-TPH2 antibody [EPR25100-29] (ab288067)

All lanes : Anti-TPH2 antibody [EPR25100-29] (ab288067) at 1/1000 dilution

Lane 1: Human brain tissue lysate

Lane 2: Human kidney tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

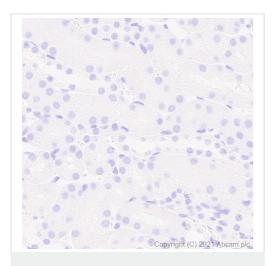
Predicted band size: 56 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST

Negative control: kidney (PMID: 12511643).

This blot was developed using a higher sensitivity ECL substrate.

Exposure time: 3 minutes

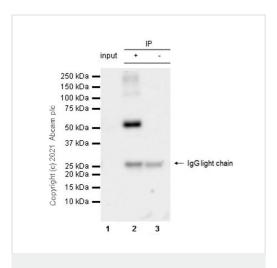


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded Mouse kidney tissue labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). No staining in mouse kidney.The section was incubated with ab288067 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) .

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunoprecipitation - Anti-TPH2 antibody [EPR25100-29] (ab288067)

TPH2 was immunoprecipitated from 0.35 mg Mouse midbrain tissue lysate 10 ug with ab288067 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab288067 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

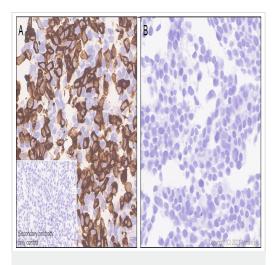
Lane 1: Mouse midbrain tissue lysate 10 ug

Lane 2: ab288067 IP in Mouse midbrain tissue lysate

Lane 3:Rabbit monoclonal lgG (ab172730) instead of ab288067 in mouse midbrain tissue lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 32 seconds

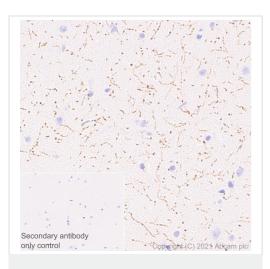


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded (A) HEK-293T cells labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Cytoplasmic staining on (A) HEK-293T cells transfected with a TPH2 expression vector containing a his tag. No staining on (B) HEK-293T cells transfected with empty vector containing a his tag. The section was incubated with ab288067 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) .

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

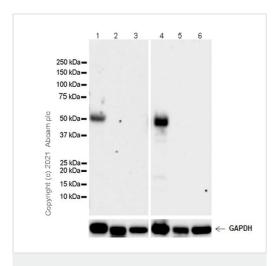


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in neural fibers of rat cerebrum. The section was incubated with ab288067 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) .

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Western blot - Anti-TPH2 antibody [EPR25100-29] (ab288067)

All lanes : Anti-TPH2 antibody [EPR25100-29] (ab288067) at 1/1000 dilution

Lane 1: Mouse midbrain tissue lysate

Lane 2: Mouse kidney tissue lysate

Lane 3: Mouse spleen tissue lysate

Lane 4: Rat midbrain tissue lysate

Lane 5: Rat kidney tissue lysate

Lane 6: Rat spleen tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 56 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST

Negative control: kidney, spleen (PMID: 12511643).

Exposure time: Lanes 1-3: 70 seconds; Lanes 4-6: 26 seconds

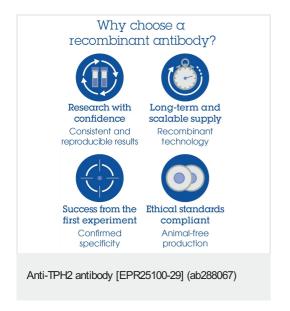
Secondary antibody only control

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TPH2 antibody
[EPR25100-29] (ab288067)

Immunohistochemical analysis of paraffin-embedded Mouse cerebrum tissue labelling TPH2 with ab288067 at 1/2000 (0.315 ug/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) . Positive staining in neural fibers of mouse cerebrum. The section was incubated with ab288067 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



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